POTASSIUM AND RIBONUCLEIC ACIDS IN HUMAN SKIN

Study of the Action of Hydrocortisone on the Psoriatic Lesion*

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We were led to undertake the studies described in this paper after a consideration of the following experimental and clinical facts:

1. There seems to be a relationship between the accumulation of potassium within the cells and the polymerization of ribonucleic acids (R.N.A.). Various experiments which have been carried out primarily on isolated organs artifically kept alive, have shown indeed that ribonuclease (20, 11), a rise of the surrounding temperature (22) reduce, both simultaneously and significantly, the concentration of cellular potassium and R.N.A. The variations of sodium, deoxy-ribonucleic acid (D.N.A.) and water are slight or nonexistent under the same conditions.

2. The corticosteroids which reduce the amount of potassium contained in the cells (9, 19) also reduce their amount in R.N.A. (3, 12, 23, 24).

3. In the course of previous work (10) it was verified that psoriatic lesions contain increased amounts of potassium (4); after treatment, clinical improvement is followed by a return to the usual potassium level. On the other hand, during clinical improvement, a decrease in plasma potassium (6, 10) can be observed. Finally, several workers have noted an increased concentration of R.N.A. in the psoriatic lesion (2, 17, 26).

It seems as though no relationship has yet been established between the accumulation of potassium and that of R.N.A. in psoriasis lesions. This accounts for our attempt to define the following points:

1) Is there any connection, in the psoriatic lesion, between the accumulation of potassium and the increase in R.N.A.?

2) What are the changes which occur after treatment by various methods (for instance, after local injection of hydrocortisone (15, 16))?

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MATERIAL AND METHODS

1. *Subjects*

Twenty-two subjects were studied (15 psoriatics and 7 controls).

The psoriatic subjects were not selected. Among them were 8 men between the age of 17 and 60, who had been affected with psoriasis for various lengths of time (from 1 to 18 years), and 7 women, between the age of 14 and 67, who had been affected with psoriasis, from 2 to 20 years. The clinical aspect of these patients was characteristic: mostly they displayed very distinct psoriatic plaques, more or less extensive, and psoriasis "guttata". Thirteen subjects had been hospitalized due to the severity and the extent of their lesions.

The 7 control subjects (4 men and 3 women) had in the past presented skin lesions on the face or the upper limbs (1 case of photo-sensitization, 1 case of "lichen planus", 5 cases of contact dermatitis); their skin was clear (not diseased) at the time of the last medical examination. Moreover, they had never presented any skin lesion in the gluteal region where skin specimens were taken for examination. They can therefore be considered as suitable control subjects.

2. *Technic and Treatment*

Forty-eight hours before the skin specimens were taken, each subject, whether psoriatic or control, was given several local intradermal injections of hydrocortisone acetate (a preparation containing 25 mg of this substance in suspension in isotonic saline). Those injections were given both into sound skin and into the lesions, in the amount of 5 mg of hydrocortisone for each injection site, the total dose being not more than 25 mg for each subject.

Intradermal injections of isotonic saline were given to the control sites in order to eliminate possible modifications due to the solvent or to trauma.

For each subject, whether psoriatic or control, skin specimens were taken simultaneously from sound skin and from skin that had been treated with hydrocortisone. Besides, in the case of psoriatic subjects, skin specimens were taken either from a lesion that had not been treated (the scales were eliminated as much as possible by

successive brushings), or from a lesion that has been treated and improved by local injections of hydrocortisone or by energetic local treatment (eosin, ultraviolet rays, 5 per cent salicylated vaseline, tar, strong reducing agents, etc.).

The skin specimens were obtained by punch-biopsy, without anesthesia, after thorough cleansing of the integument with alcohol and ether. They were always localized around the upper gluteal region. The skin specimens thus removed are 3 mm in diameter, their wet weight is about 10 to 15 mg.

3. *Chemical Methods*

For determination of potassium, sodium and water, the skin specimens were weighed when wet, and then handled following the methods of Lowry and Hastings (7). The amount of potassium and sodium in the extraction fluid were determined by flame photometry. The skin specimens were then dried and weighed a second time. Tissue hydration was calculated on the basis of wet weight and dry weight of the same specimen.

For determination of R.N.A. and D.N.A., the skin specimens were treated according to the technic of Ogur and Rosen modified by Steinert (18).

Thus we have the possibility of comparing, for each subject, the concentrations in potassium, sodium, water, R.N.A. and D.N.A.:

1. in normal skin, whether treated or not by hydrocortisone;

2. in normal skin and in psoriatic skin, before and during clinical improvement.

RESULTS

Results are given in table I; all comparisons of averages were submitted to statistical control ("t" test of Student-Fischer).

TABLE I

		Control Subjects		Psoriatic Subjects				
		Sound skin	Sound skin treated with hydro-cortisone	Sound skin	Sound skin treated with hydro-cortisone	Psoriatic lesion	Psoriatic lesion treated	
							with hydro-cortisone	with local ointments
Potassium μEq/g fresh weight	m	*21, 9*	*19, 7*	*23, 8*	*19, 1*	*46*	*29, 7*	*26, 5*
	n	14	9	II	9	10	6	10
	m	0, 61	0, 54	0, 73	0, 74	1, 70	0, 52	2, 20
Potassium μEq/g defatted dry weight	m	*59, 7*	*51, 8*	*64, 6*	*49, 2*	*126, 3*	*81*	*64, 5*
	n	14	9	II	9	10	6	10
	m	1, 66	1, 48	1, 87	1, 88	4, 41	1, 41	5, 55
Sodium μEq/g fresh weight	m	*104, 2*	*111, 5*	*109, 1*	*112, 6*	*119, 5*	*109, 8*	*114*
	n	14	9	II	9	10	6	6
	m	2, 11	2, 65	2, 35	3, 18	2, 72	2, 16	2, 15
Water per cent of fresh weight	m	*63, 5*	*62*	*61, 4*	*61, 2*	*63, 6*	*63*	*60, 8*
	n	8	8	10	8	8	6	6
	m	0, 43	0, 41	0, 78	0, 86	0, 81	0, 68	0, 59
R.N.A. μMol of P per g defatted dry weight	m	*24, 1*	*21*	*26, 8*	*22, 8*	*52, 1*	*38, 4*	*37, 5*
	n	9	10	21	17	9	10	5
	m	0, 75	0, 83	0, 99	0, 83	2, 22	2, 18	2, 31
D.N.A. μMol of P per g defatted dry weight	m	*57, 7*	*62*	*54, 7*	*57, 1*	*58, 5*	*65, 7*	*58, 2*
	n	9	9	22	17	9	10	5
	m	2, 67	2, 10	1, 65	2, 17	2, 31	2, 37	3, 55
Quotient R.N.A./D.N.A.	m	*0, 42*	*0, 34*	*0, 49*	*0, 40*	*0, 89*	*0, 58*	*0, 64*

m = mean; n = number of samples; m = standard error of the mean.

1. The sound skin of control subjects does not differ from the sound skin of psoriatic subjects with regard to the average contents in potassium, sodium, water, R.N.A. and D.N.A. Nevertheless, it should be noted that, in 5 cases out of 15, a comparison between the individual values reveals that the normal skin of the psoriatic subjects shows a slight increase in potassium and R.N.A., which accounts for the differences between the averages just considered.

2. The psoriatic lesion, when not treated, is particularly rich in potassium and R.N.A.: the values are twice as high as those which characterize normal skin. Such variations are therefore important and were found in each of the 8 cases we examined.

The larger amount of sodium in the psoriatic lesion, as compared to normal skin, is of doubtful significance.

As far as water and D.N.A. are concerned, the averages do not differ from each other. (The concentrations in D.N.A. of psoriatic lesions are normal in 5 cases, higher in 2 cases, and lower than the average in one case.)

3. A local intradermal injection into normal skin given to 14 psoriatic subjects and 6 control subjects, causes after 48 hours a significant decrease in potassium and R.N.A. The concurrent variations in sodium, water and D.N.A. are not significant.

A detailed study of the above results shows a decrease in potassium and R.N.A. for 15 subjects, but an increase in potassium and R.N.A. for 2 subjects, and no variation for the other 3.

4. It can be seen that the potassium and R.N.A. content of the lesions which have been improved by treatment are far lower than that of the lesions which have not been treated, though these amounts remain higher than in the case of sound skin. Sodium and water show no significant variations.

The decrease in potassium and R.N.A. of a psoriatic lesion during healing cannot be accounted for by the type of treatment that was given, since such variations can be observed in the 4 cases where an improvement was reached through applying local treatments, as well as in the 8 cases where the improvement followed local injections of hydrocortisone.

There appears a small difference as far as the values of D.N.A. are concerned. The amount of D.N.A. in psoriatic lesions, in lesions which have been improved by local treatment, and in normal skin do not differ whereas there is a greater amount of D.N.A. in the lesions which have been treated with hydrocortisone. Thus the R.N.A./D.N.A. quotient decreases in normal skin as well as in psoriatic skin following local injection of hydrocortisone.

DISCUSSION

We fully realize that our biopsy specimens consist of both epidermis and dermis and that such material may contain relatively more epidermis when taken from lesions due to diseases which cause acanthosis. The histology of a lesion is always to be taken into consideration when evaluating biochemical results obtained on such lesions. Thus it is not surprising that, more particularly in psoriasis, where there is both acanthosis and parakeratosis with presence of increased amounts of nuclear materials, we were able to find increased amounts of R.N.A. and K. It is not surprising either that when the skin resumes its normal clinical and histological appearance these findings tend to revert to "normal".

It is however remarkable that, in the psoriatic lesions, the levels of D.N.A. did not rise. Despite increased epidermal cellularity and increased amounts of nuclear material evidenced histologically, we did not find an increase in the concentration of D.N.A. in line with the increase in the amount of R.N.A. This would have been expected because of the presence of increased nuclear material.* The reasons for our findings are not clear at the present time. Brachet (1) has noted that" . . . all organs which synthesize large amounts of proteins, whether for growth or multiplication, are always rich in R.N.A. which is localized in the nucleolus and the cytoplasm. . . ."

The above results seem to confirm two important facts: in psoriatic lesions, potassium and R.N.A. values are increased; sodium, water, and D.N.A., show no, or very slight, variation. There seems to be a relationship between the increase in potassium and that in R.N.A. Indeed, the variations of these two constituents occur in the same direction (increase or decrease) and are

* Dr. G. K. Steigleder (personal communication) has noted that nuclei in parakeratotic layers do not stain like normal DNA. They remain Feulgen positive, however, this may indicate an alteration in the structure of DNA in parakeratotic nuclei; the chemical method which we used for DNA determination may not reveal this abnormal material.

simultaneous. It seems as though the factors which modify the concentrations of the tissues (sound skin, psoriatic lesions, isolated hearts, etc.) in R.N.A., also modify their concentration in potassium. We know also that the presence of potassium (and magnesium, the concentration of which rises in the psoriatic lesion (2)) seems to be necessary to the synthesis of proteins and R.N.A. (20).

The accumulation of R.N.A. and potassium might be related to the process of protein synthesis (1, 3, 5, 13, 14, 17) and to the high concentration of pentoses in psoriatic lesions (5, 26).

Yet, it does not look as though the accumulation of potassium and R.N.A. were a phenomenon characteristic of psoriasis. It might be only an indication of a process of active synthesis taking place in the cells. Studies now in progress tend to show that lesions due to eczema (contact dermatitis) have a rather high content in R.N.A. and in potassium.

The clinical improvement of psoriasis is attended by a considerable decrease in R.N.A. and in potassium, whatever be the therapeutic methods. Such a result occurs after a local injection of hydrocortisone, or after local application of various drugs. The action of hydrocortisone does not seem to be specific; this substance also causes a decrease in R.N.A. and potassium in sound skin.

A comparison should be made between the diminished concentration in R.N.A. of the healed psoriatic lesions and the results which have been obtained after treatment with aminopterin, an antimetabolite which interferes with the synthesis of nucleic acids (8).

The reasons why D.N.A. did not increase concommitantly with, or even to a greater extent than, R.N.A. in psoriatic lesions are not clear and should be investigated.

SUMMARY

The authors measured the amounts of potassium, sodium, R.N.A. and D.N.A. in the skin of the gluteal region of 22 subjects: 15 psoriatics and 7 controls. The skin specimens were taken by means of a punch-biopsy, 1) from sound skin and from sound skin treated with hydrocortisone (local injection); 2) from psoriatic lesions which had not been treated and from psoriatic lesions which had been treated with local injection of hydrocortisone or with the usual local treatments. Insofar as potassium, sodium, R.N.A., and D.N.A. are concerned:

1. The sound skin of the psoriatic subjects, in 10 cases out of 15, is not different from the sound skin of the control subjects.

2. The psoriatic lesion is particularly rich in R.N.A. and in potassium; its concentration in sodium, water and D.N.A. is close to that of sound skin. The reasons for the lack of increase in D.N.A. in the psoriatic lesions are not known.

3. Whatever the treatment given, clinical improvement of the psoriatic lesion is attended by a decrease in R.N.A. and in potassium.

4. After 48 hours, the local injection of hydrocortisone acetate causes a significant decrease in the concentrations of R.N.A. and potassium of sound skin, and even more so in psoriatic lesions; there is a small increase in the amount of D.N.A.

5. There seems to be a relation between the concentrations in R.N.A., and in potassium of the skin.

6. The accumulation of R.N.A. and potassium in the psoriatic lesion does not seem specific.

7. The improvement of psoriatic lesions which results from the local injections of hydrocortisone is not specific either, but should be attributed to the rather wide action of corticosteroids, which has been observed on several tissues.

REFERENCES

1. Brachet, J.: Biochemical cytology. Academic Press, New York, 1957.
2. Braun Falco, O.: The histochemistry of psoriasis. Ann. N. Y. Acad. Sc., **73:** 936, 1958.
3. Chargaff, E. and Davidson, J. N.: The Nucleic Acids, Volume 2. Academic Press, New York, 1955.
4. Dörffel, J.: Klinische experimentelle und chemische studien über den Einfluss der Ernahrung auf Entzundungs vorgange in der gesunden und krander Haut. Arch. f. Dermat. u. Syph., **162:** 621, 1930.
5. Flesch, P.: Chemical data on human epidermal keratinization and differentiation. J. Invest. Derm., **31:** 63, 1958.
6. Levy, R., Reinberg, A., Sidi, E. and Hincky, M.: Diminution du potassium plasmatique lors de l'amélioration clinique du psoriasis. Sem. Hôp. Paris, **31:** no. 34, 1955.
7. Lowry, O. H. and Hastings, A. B.: Histochemical changes associated with ageing. Methods and calculations. J. Biol. Chem., **143:** 257, 1942.
8. Rees, R. B. and Bennett, J. H.: Further observations on aminopterin for psoriasis. J. Invest. Derm., **32:** 61, 1959.
9. Reinberg, A. and Stolkowski, J.: Influence de la désoxycorticostérone sur le potassium cellulaire. Ann. Endocr. (Par.) **13:** 559, 1952.
10. Reinberg, A., Bourgeois-Spinasse, J., Hincky, M. and Sidi, E.: Metabolic studies

in psoriasis—Skin potassium and plasma potassium, chlorid and alkaline values during clinical improvement. J. Invest. Derm., **31:** 231, 1958.

11. Reinberg, A. and Stolkowski, J.: Influence de la polymérisation des acides ribonucléiques sur les échanges en potassium d'organes isolés. Arch. Sc. Physiol. (Par.), **12:** 293, 1958.
12. Reinberg, A. and Stolkowski, J.: Action des corticosteroides sur le potassium des cellules: rôle possible des acides ribonucléiques (A.R.N.), Ier Congres International d'Endocrinologie. Copenhague, 1960. Excerpta Medical Edit. and Ann. Endocr. (Par.), **21:** 715, 1960.
13. Rothman, S.: Physiology and Biochemistry of the Skin, Chicago, University of Chicago Press, 1954.
14. Schmidli, B. and Paschoud, J. M.: Acides aminés et polypeptides dans la peau atteinte de psoriasis. Dermatologica, **110:** 323, 1955.
15. Sidi, E. and Bourgeois-Gavardin, J.: Modifications cutanées consécutives aux applications et aux injections locales d'acétate d'hydrocortisone. Presse Med. **82:** 1760, 1953.
16. Sidi, E. and Bourgeois-Spinasse, J.: L'hydrocortisone en dermatologie. I vol. 73 p. Lab. Roussel Edit. Paris, 1956.
17. Steigleder, G. K.: Zur Funktion der Acanthose. Arch. f. Dermat. u. Syph., **200:** 377, 1955.
18. Steinert, M.: La synthese de l'acide ribonucleique au cours du développement embryonnaire des Batraciens. Bull. Soc. Chim. Biol. (Par.), **33:** 549, 1951.
19. Stolkowski, J. and Reinberg, A.: Inhibition de l'entrée du potassium dans les cellules sous l'action de la cortisone et de la désoxycorticostérone. Ann. Endocr. (Par.), **13:** 947, 1952.
20. Stolkowski, J. and Reinberg, A.: Action de la ribonucléase sur les échanges ioniques (potassium) d'organes isolés. Comptes rendus. Acad. Sc., **246:** 2420, 1958.
21. Stolkowski, J. and Reinberg, A.: Influence de la pression osmotique du milieu sur les mouvements du potassium et la polymérisation des acides ribonucleiques de cellules animales, Arch. Sci. Physiol. (Par.), **14:** 25, 1960.
22. Stolkowski, J., Reinberg, A. and Blanc-Dingeon, F.: Action de la température sur les mouvements du potassium et la polymérisation des acides ribonucleiques de cours isolés d'*Helix aspersa* (Müller) maintenus en survie. J. Physiol. (Par.), **52:** 228, 1960.
23. Tremolieres, J., Derache, R. and Griffaton, G.: Effets de la cortisone sur les nucleoproteines du foie et du muscle. Ann. Endocr. (Par.) **15:** 827, 1954.
24. Tremolieres, J., Derache, R. and Griffaton, G.: Effets de la cortisone et de la surrénalectomie sur les variations conjointes du sodium et du potassium et de quelques fractions phosphorées dans le foie et le muscle du rat. Ann. Endocr. (Par.), **15:** 836, 1954.
25. Webster, G. C.: Effects of nucleosides and their derivatives in protein synthesis in cell-free extracts of pea roots. Fed. Proc., **15:** 380, 1956.
26. Wheatley, V. R. and Farber, E. M.: Studies on the chemical composition of psoriatic scales. J. Invest. Dermat., **36:** 199, 1961.